

## **1. Summary**

**Project Title:** Egg parasitism of the Virginia creeper (*Erythroneura ziczac*), a newly invasive leafhopper pest in California (Year 2)

**Principal Investigators:** Kent Daane, Glenn McGourty, Serguei Triapitsyn, Lucia Varela

### **Summary:**

Organic grape growers in Mendocino and Lake County have been experiencing severe outbreaks of the Virginia creeper leafhopper (*Erythroneura ziczac*) for the past 3 years. Feeding by *E. ziczac* causes leaf stippling and reduced photosynthesis which can impact crop yield and quality. The primary natural enemies of *E. ziczac* are the small ‘mymarid’ egg parasitoids *Anagrus daanei* and *Anagrus tretiakovae*. A related pest, the Western grape leafhopper (*Erythroneura elegantula*) is also parasitized by *A. daanei* as well as *Anagrus erythroneurae*. *Erythroneura ziczac* and *E. elegantula* are commonly found together in North Coast vineyards. *Anagrus daanei* is the parasitoid species of most importance for *E. ziczac* control, whereas *A. tretiakovae* is rarely found in California.

Our approach to improving *E. ziczac* control involves a combination of short- and long-term strategies. Short-term work focuses on the evaluation of Organic Materials Review Institute (OMRI) approved pesticides. In 2014, we tested Stylet oil and DeBug® Turbo (applied twice) and Pyganic® (applied once) on the development of the first leafhopper brood. All of these products significantly reduced *E. ziczac* nymph populations relative to an untreated control.

Long-term strategies are focused on the identification and evaluation of *Anagrus* parasitoids to improve biological control. A survey in Mendocino, Lake, Napa (Pope Valley), Yolo and El Dorado County vineyards found that *E. ziczac* parasitism was consistently low (0-2%) with the exception of Yolo County, where rates reached 10-15%. Surprisingly, *A. daanei* was attacking *E. elegantula* in all of the surveyed vineyards, but only in Yolo County was it attacking both *E. elegantula* and *E. ziczac*. Therefore, we questioned whether or not the *A. daanei* in Yolo County are the same species as the *A. daanei* that don’t attack *E. ziczac* in other regions. Molecular comparison of the *A. daanei* from different Californai regions is still in progress, but to date morphological evaluations have not shown any differences among the *A. daanei* populations tested. We conducted a trial in which we forced *A. daanei* from Mendocino County onto *E. ziczac* eggs in order to see whether or not, in the absence of their preferred *E. elegantula* host, they would attack the *E. ziczac* eggs. Findings from this study indicated they would not.

In another trial, we separately inoculated potted grape vines with *E. ziczac* eggs from Mendocino, Lake and Yolo County and then exposed sets of these vines to the *A. daanei* in each of these regions. Results showed fairly consistent parasitism of all three *E. ziczac* populations by the *A. daanei* in Yolo County. Having verified that the *A. daanei* in Yolo County will readily attack the *E. ziczac* population in Mendocino and Lake County, we now feel that there is adequate evidence to support a collection and re-release program in which *A. daanei* from Yolo County are introduced into Mendocino and Lake County vineyards. This redistribution of California parasitoid material would be carried out in conjunction with an area-wide IPM program to promote additional best management practices to further reduce *E. ziczac* outbreaks.

## **2. Annual or Final Report**

This is an annual report (year 2)

## **3. Project Title and UGMVE proposal number**

“Egg parasitism of the Virginia creeper (*Erythroneura ziczac*), a newly invasive leafhopper pest in California.” (Proposal #2015-1493)

## **4. Principle Investigator/Cooperator(s):**

### **Principal Investigators:**

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## **5. Objective(s) and Experiments Conducted to Meet Stated Objective(s):**

The 2014 project objectives were modified from the originally proposal because this project was awarded \$27,000 of the requested \$54,818. Our modified objectives were based on an assessment of project priorities that would be of most direct benefit to growers.

### *1. Evaluate the efficacy of materials for Western grape leafhopper control on Virginia creeper leafhoppers.*

We established a trial in a Cabernet Sauvignon vineyard in Mendocino County to test the efficacy of three OMRI-approved products for *E. ziczac* control that are commonly used against *E. elegantula*. There were four treatments: Stylet oil (1% solution), DeBug® Turbo (32 fl. oz. per acre), Pyganic® (13.5 fl. oz. per acre) and a no treatment control. All solutions were acidified to a pH of 5.5 with 1 qt./acre BioLink 3-3-3 fertilizer and 1 qt./acre Biolink Cal Plus. Treatments were replicated three times in a randomized complete block design. Each replicate (plot) was six vine rows and data was collected from the middle two rows of each plot. Stylet oil and DeBug® Turbo were applied on May 23 and June 9. Pyganic® was applied on June 9. The control was untreated. Products were applied with a speed sprayer at 150 gallons/acre. A pre-count of *E. ziczac* nymphs was made on May 22. Nymph counts after the first insecticide application (May

23) were made on May 30 and June 6. Nymph counts after the second insecticide application (June 9) were made on June 12. The number of leafhopper nymphs per leaf was counted on 20 leaves per plot selected from each of 20 random vines in the center two rows of each plot. We selected leaves with stippling damage. Treatment effects on leafhopper nymph counts were analyzed with ANOVA and Tukey's HSD procedure was used to detect differences between individual treatments.

## 2. Manipulate parasitoid populations in order to improve biological control of Virginia creeper leafhopper in California.

### 2a. Northern California survey of *E. ziczac* parasitism

In 2013, surveys in Mendocino County of *E. ziczac* parasitism revealed that although the key parasitoid *A. daanei* is present in the county, it is only attacking *E. elegantula* (western grape leafhopper). In 2014, an expanded survey was conducted across northern California in order to better contextualize the pest-parasitoid dynamics observed in Mendocino County the previous year. The survey was carried out in May-July 2014 and included vineyards in Mendocino, Lake, Napa (Pope Valley), Yolo and El Dorado County. In each county 4-8 vineyard sites were surveyed. At each site, 10-30 leaves were collected and evaluated in the laboratory for parasitism of both *E. ziczac* and *E. elegantula*. Eggs of both leafhopper species that clearly contained a developing *Anagrus* wasp were isolated, adult wasps reared out and collected, stored in alcohol, and sent to Dr. Serguei Triapitsyn for identification.

### 2b. *Anagrus daanei* no-choice tests

In a greenhouse trial, potted grape vines were placed into cages and inoculated with adults of either *E. elegantula* or *E. ziczac* that were collected from Mendocino County. We then reared *A. daanei* from *E. elegantula* eggs (also collected in Mendocino County) and introduced the wasps onto the caged vines. The goal of this study was to evaluate whether or not *A. daanei* from Mendocino County would parasitize *E. ziczac* eggs when faced with no other choice of viable host.

This trial had two treatments: (1) vines inoculated with *E. elegantula* and (2) vines inoculated with *E. ziczac*. Each treatment was replicated nine times (18 inoculated, caged vines total). For each replicate, a single potted grape vine (Cabernet Sauvignon 06) with five fully expanded mature leaves was placed into an observation cage (46 x 44 x 55 cm.). The cage was then randomly assigned to either the "*E. elegantula*" or "*E. ziczac*" treatment. Vines were accordingly inoculated with approximately 60-80 adult leafhoppers (1:2 M:F) that were collected from a vineyard in Mendocino County earlier in the day (August 8). The inoculated, caged vines were then placed in the greenhouse and held at  $71.8 \pm 0.1^\circ\text{F}$  and  $71.8 \pm 0.1\%$  RH.

In order to rear the *A. daanei* used in this trial, grape leaves were collected from a vineyard in Mendocino County and placed into emergence chambers on the same day that we collected the adult leafhoppers (August 8). Each emergence chamber received 50 leaves and was randomly assigned to one of the caged vine replicates (i.e. each of the inoculated, caged vines would receive all wasps from one emergence chamber). Emergence chambers were held at  $86.1 \pm 0.2^\circ\text{F}$  and  $41.8 \pm 0.2\%$  RH. For the next eleven days (August 9-19) the chambers were checked three times a day (8am, 12pm, 4pm) for emerged *Anagrus* wasps. When an *Anagrus* wasp was

found, it was isolated and immediately introduced onto its assigned caged vine. The *Anagrus* wasps emerged at different rates, but within two days at least one female wasp had been introduced onto each caged vine. Each caged vine ultimately received an average of  $29.9 \pm 6.4$  wasps ( $24 \pm 5.3$  females,  $5.9 \pm 1.4$  males). An average of  $25.6 \pm 4.1$  females were introduced into each “*E. ziczac*” replicate and an average of  $23.8 \pm 3.8$  females introduced into each “*E. elegantula*” replicate.

The grape leaves came from a vineyard that had a mixed population of *E. elegantula* and *E. ziczac*. Previous surveys had shown that only *E. elegantula* eggs were being parasitized in this region, primarily by *A. daanei* and *A. erythroneurae*. To verify that only the *E. elegantula* eggs were parasitized on these leaves, a subset of leaves (150 leaves) were inspected under a dissecting microscope. Additionally, all *E. elegantula* eggs that were found to be parasitized by an *Anagrus* wasp were isolated and reared out for identification. We reared a total of 13 wasps in this manner from *E. elegantula* eggs. While we did find two *E. ziczac* eggs that appeared to have been parasitized, this represents  $<0.001\%$  of the total eggs evaluated and likely did not translate into any significant amount of error in this experiment.

Additionally, *Anagrus* wasps are so small that it is difficult to differentiate species when handling them live. Since *E. elegantula* eggs can be parasitized by either *A. daanei* or *A. erythroneurae*, a subset of *Anagrus* wasps were collected from each of the emergence chambers in order to verify what species were being introduced into the cages. We collected a total of 79 wasps for identification ( $4.4 \pm 0.7$  female wasps per emergence chamber).

After 11 days no further *Anagrus* wasps were introduced onto the caged vines. The vines were held in the greenhouse for an additional two weeks (August 19 – September 1) in order to allow any parasitized leafhopper eggs to fully develop. On September 1 leaves from each of the caged vines were removed and evaluated for parasitism in the laboratory. Any leafhopper eggs that contained a developing *Anagrus* wasp were isolated in order to rear out the parasitoid for identification.

### 2c. *Erythroneura ziczac* cross-exposure experiment

The goal of this experiment was to determine whether or not the *A. daanei* in Yolo County would attack *E. ziczac* eggs from Mendocino and Lake County. To do this, separate sets of potted grape vines were inoculated with *E. ziczac* eggs from either Mendocino, Lake or Yolo County and were then exposed to the *Anagrus* parasitoids in Mendocino, Lake and Yolo County.

Adults of *E. ziczac* were collected from vineyards in Mendocino, Lake and Yolo County (July 6). A set of 21 potted grape vines (Cabernet Sauvignon 06) was then separately inoculated with leafhoppers from each of the three counties (21 vines/county x 3 counties = 63 vines total). Each vine had four healthy mature leaves and was inoculated by placing a small clip-cage (4 cm. diameter) on each of the four leaves and introducing 10-15 *E. ziczac* adults (1:1 M:F) into each clip-cage (1 clip-cage per leaf x 10-15 adults per cage x 4 leaves per vine = 40-60 adults per potted vine). A 5-gallon paint strainer bag (Supertuff® Paint Strainers, Trimaco, Morrisville, NC) was placed over each of the potted vines in order to further protect against any cross-contamination. Vines were held in the greenhouse ( $71.3 \pm 0.2^\circ\text{F}$  and  $62.5 \pm 0.3\%$  RH) for 48 hours (July 6-8) in order to allow *E. ziczac* to oviposit into the leaves.

On July 9 the paint strainer bags, clip-cages and adult *E. ziczac* were all removed and the vines were brought to field sites for exposure to the resident *Anagrus*. There was one field site in

each of the three counties. There were seven replicates at each field site. Replicate plots consisted of one vine row with a one row buffer between plots (15 vine rows total); each row contained 50-100 grape vines. One potted vine inoculated with *E. ziczac* from each of the three *E. ziczac* populations (Mendocino, Lake and Yolo) was randomly placed between vines 10-50 in each of the row-plot replicates. None of the potted vines were placed at the first or last ten vines in any row. All of the potted vines were provided with water by placing them in a 2-gallon bucket filled with approximately 0.5 gallons of water.

The potted vines were left in the field for 6 days (July 9-15). Vines were then retrieved from the field sites and brought back to the greenhouse to rear out any parasitoids that may have attacked the *E. ziczac* eggs. This was done by placing a clip-cage back onto each leaf covering the area where the eggs had initially been deposited. The vines were then held in the greenhouse ( $71.3 \pm 0.2^\circ\text{F}$  and  $62.5 \pm 0.3\% \text{RH}$ ) for 13 days (July 15-28) to allow any parasitoids to develop and emerge. Each clip-cage contained a small strip of a sticky-card (Seabright Laboratories, Emeryville, CA) to capture the emerging parasitoids. All of the potted vines were watered daily in the greenhouse.

On July 28 leaves were removed from the potted vines and evaluated for parasitism. The sticky-card inside of each clip-cage was also inspected for parasitoids. Orange oil (Histo-clear®, National Diagnostics, Atlanta, GA) was used to remove any parasitoids from the sticky-card, which were then preserved in alcohol and sent to Dr. Serguei Triapitsyn for identification.

Linear mixed-effect models were used to evaluate *E. ziczac* parasitism rates. Fixed effects included “total eggs per vine”, “exposure site”, “leafhopper population”. “Replicate” was used as a random effect. Single-term deletion tests were then used to determine the significance of fixed effects.

## **6. Summary of Major Research Accomplishments and Results by Objectives**

### *1. Evaluate the efficacy of materials for Western grape leafhopper control on Virginia creeper leafhoppers.*

Before insecticide treatments were applied, the numbers of nymphs were not statistically different among plots. After the first treatment, *E. ziczac* populations decreased to 2.6 and 5.5 nymphs/leaf in the Stylet oil and DeBug® Turbo treatments, respectively (Figure 1, Table 1), and both treatments were lower than the no spray control (p-value < 0.001). Pyganic® had not yet been sprayed. The evaluation on June 6 found similar nymph numbers in the Stylet oil and DeBug® Turbo treatments (Figure 2, Table 1), which were again significantly lower than the no spray control. After the second spray, which now included Pyganic®, the numbers of nymphs in all spray treatments were lower than the control (Figure 3, Table 1; p-value < 0.001)

All of the spray treatments were applied during the development of the first brood. The first treatments of Stylet oil and DeBug® Turbo were applied when 1<sup>st</sup> and 2<sup>nd</sup> instar nymphs were present. Both treatments significantly reduced the number of nymphs per leaf compared to the untreated control, with Stylet oil having a significant lower nymph count than DeBug® Turbo on the first evaluation (May 30). After the treatment on May 23, evaluations were conducted 7 and 14 days after treatment to determine if on the DeBug® Turbo treatment we would observe a decrease in population through time. DeBug® Turbo has the active ingredient azadirachtin which is an insect growth regulator. We did not observe a decrease but an increase

between the evaluation at 7 and 14 days in both the DeBug® Turbo and Stylet oil treatments. This increase may be due to the hatching of new eggs after treatment.

When the application was done on June 9 all nymph stages (1<sup>st</sup> to 5<sup>th</sup> instar) were present. The two treatments that had previously received an application on May 23 had all nymphal stages present, however the majority of the population were younger nymphs due to the treatment. All treatments: Stylet oil applied twice, DeBug® Turbo applied twice and Pyganic® applied once had significantly lower numbers of leafhopper nymphs/leaf than the control (Figure 3). At the time of the evaluation on June 12, the 5<sup>th</sup> instar nymphs were beginning to turn into adults. On this date nymphs of the next generation had not yet started to hatch. This may explain why the number of nymphs in the untreated control was lower than in previous evaluations. Since nymphs were turning into adults and the next generation was about to begin, no further evaluations were conducted.

Table 1. Average number of leafhopper nymphs/leaf ( $\pm$ SEM). Numbers within a row followed by the same letter are not significantly different at  $\alpha=0.05$  using Tukey’s HSD procedure.

Date	Treatment			
	Stylet oil	DeBug® Turbo	Pyganic®	Control
May 22	15.9 $\pm$ 0.7 a	15.8 $\pm$ 1.1 a	15.5 $\pm$ 1.0 a	15.9 $\pm$ 0.8 a
May 23	Spray	Spray	No spray	No spray
May 30	2.6 $\pm$ 0.8 a	5.5 $\pm$ 0.8 b	15.7 $\pm$ 1.0 c	16.1 $\pm$ 1.0 c
June 6	5.6 $\pm$ 0.7 a	7.7 $\pm$ 1.1 a	16.6 $\pm$ 1.7 b	14.7 $\pm$ 1.0 b
June 9	Spray	Spray	Spray	No spray
June 12	0.6 $\pm$ 0.2 a	1.3 $\pm$ 0.5 a	0.0 $\pm$ 0.0 a	7.7 $\pm$ 0.8 b

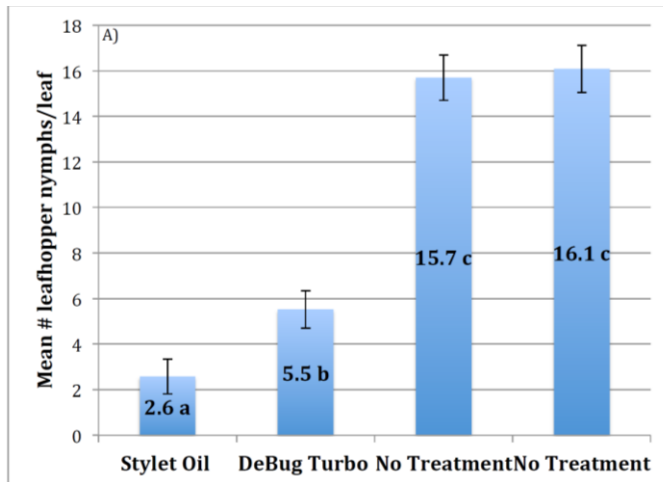


Figure 1. Leafhopper nymphs/leaf on May 30

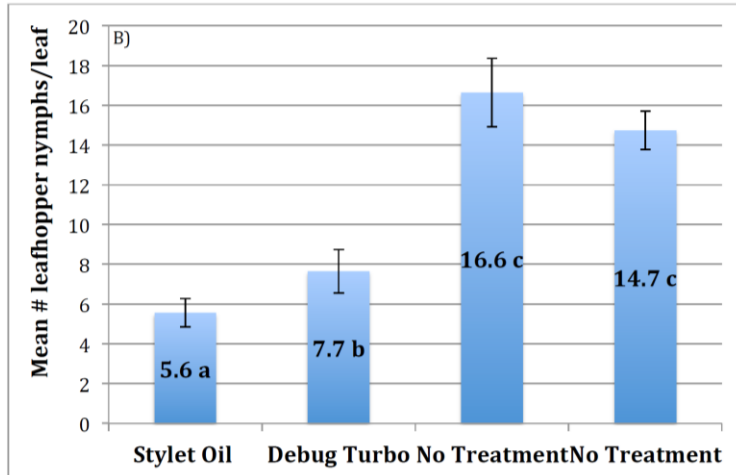


Figure 2. Leafhopper nymphs/leaf on June 6

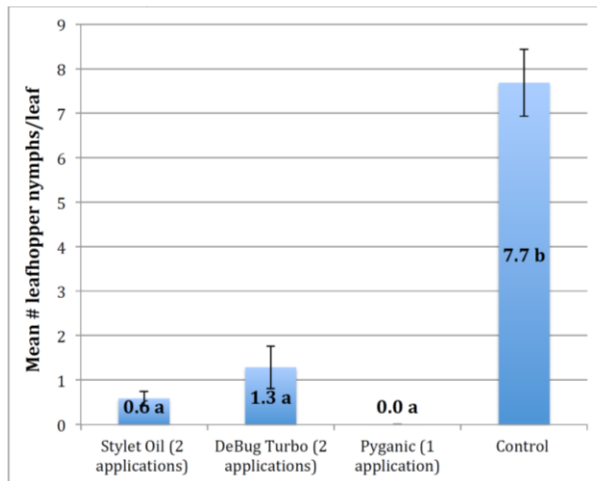


Figure 3. Leafhopper nymphs/leaf on June 12

2. Manipulate parasitoid populations in order to improve biological control of Virginia creeper leafhopper in California.

2a. Northern California survey of *E. ziczac* parasitism

The survey work in 2014 found that whereas there is effectively little to no parasitism of *E. ziczac* in Mendocino, Lake, Napa (Pope Valley) and El Dorado Counties, the *E. ziczac* in Yolo County are regularly being parasitized by *Anagrus* wasps (Table 2). *Anagrus* wasps were reared for identification from both *E. ziczac* and *E. elegantula* eggs in all of the surveyed regions. Both *A. erythroneuræ* and *A. daanei* were primarily reared from *E. elegantula* eggs whereas *A. daanei* was mostly reared from *E. ziczac* eggs (Table 2). Molecular work to compare *A. daanei* populations is still in progress, but all of the morphological comparisons to date have not found any differences that would indicate that these groups are separate species.

Table 2. Parasitism rates and *Anagrus* species reared from *E. ziczac* and *E. elegantula* in northern California vineyards.

County	<i>E. ziczac</i>		<i>E. elegantula</i>	
	Parasitism rate	<i>Anagrus</i> species	Parasitism rate	<i>Anagrus</i> species
Mendocino	<0.01%	<i>A. daanei</i>	25%	<i>A. erythroneuræ</i> <i>A. daanei</i>
Lake	-	<i>A. daanei</i> <i>A. erythroneuræ</i>	-	<i>A. daanei</i> <i>A. erythroneuræ</i>
Napa (Pope Valley)	<0.01%	-	27%	<i>A. daanei</i> <i>A. erythroneuræ</i>
Yolo	13%	<i>A. daanei</i>	40%	<i>A. daanei</i> <i>A. erythroneuræ</i> <i>A. tretiakovæ</i>
El Dorado	1%	<i>A. daanei</i>	19%	<i>A. daanei</i>

2b. *Anagrus daanei* no-choice tests

Identification of a subset of the *Anagrus* wasps used in this trial revealed that approximately 75% of the wasps introduced into the experimental cages were *A. erythroneuræ* and 25% *A. daanei* (Figure 4). There was also cross-contamination of the experimental leafhopper treatments, as we found some *E. ziczac* eggs on the potted vines in the “*E. elegantula*” treatment and *E. elegantula* eggs in the “*E. ziczac*” treatment (Figure 5).

Similar to what has been observed in the field, we saw fairly consistent parasitism of *E. elegantula* eggs but practically no parasitism of *E. ziczac* eggs (Figure 6). Where parasitism of *E. ziczac* did occur, it was found to be the result of attack by *A. erythroneuræ* rather than *A. daanei*. This is surprising because *A. erythroneuræ* is only known to attack *E. elegantula* and *E. variabilis* (Variegated leafhopper), not *E. ziczac*.

The goal of this experiment was to see whether or not *A. daanei* from the Mendocino County population could be “forced” to attack *E. ziczac* by confining it to a potted grape with only *E. ziczac* eggs. In this study there was no observed parasitism of *E. ziczac* by *A. daanei*, although it must be acknowledged that a relatively low number of these wasps were introduced into the experimental cages and, due to cross-contamination of the two leafhopper treatments, the wasps were not entirely isolated with either species of leafhopper.



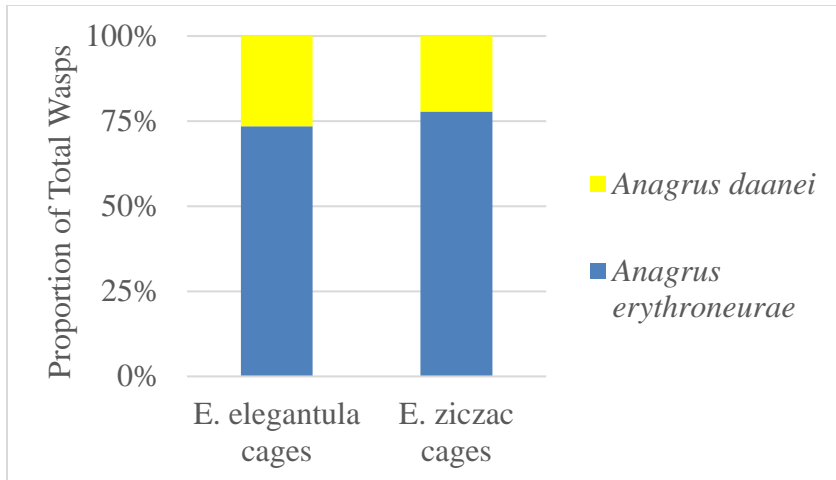


Figure 4. Relative proportion of *A. erythroneurae* and *A. daanei* introduced onto the caged vines.

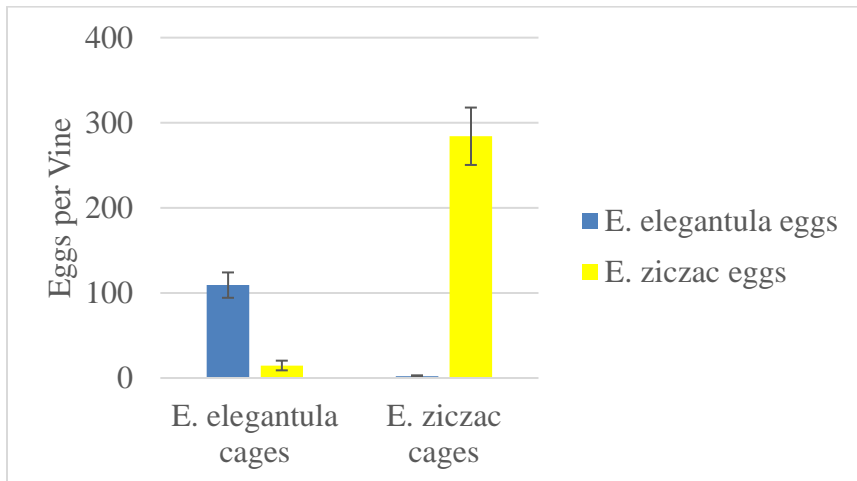


Figure 5. Average number of leafhopper eggs on potted vines in each experimental treatments.

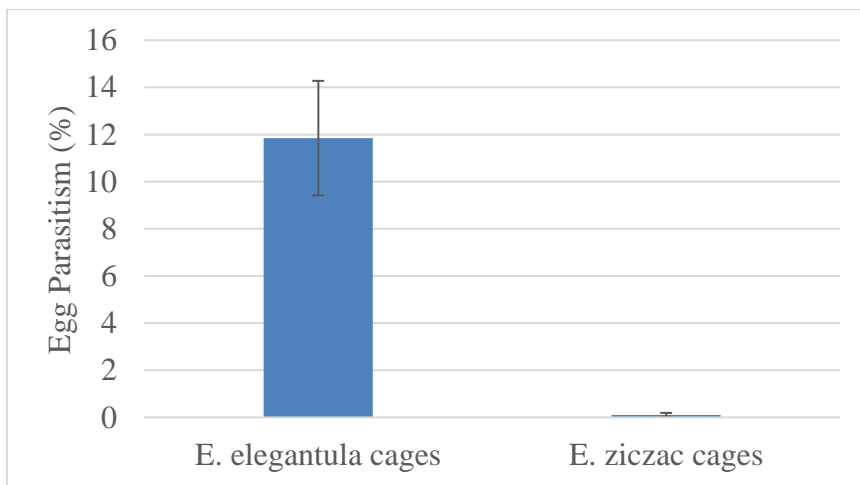


Figure 6. Leafhopper egg parasitism rates in the two treatments

2c. *Erythroneura ziczac* cross-exposure experiment

All three populations of *E. ziczac* eggs were parasitized by *Anagrus* wasps in Yolo County, but not in Mendocino or Lake County (Figure 7). Results of the statistical analysis indicated that both “exposure site” and “source population” had a significant influence on parasitism rates (Table 3) but there was no effect due to “total eggs per vine”.

The *Anagrus* species reared from the parasitized eggs were predominantly *A. daanei*, although *A. erythroneurae* and *A. tretiakovae* were also found (Figure 8). We were surprised to find both *A. erythroneurae* and *A. tretiakovae*, as the former is not known to attack *E. ziczac* and the latter is rarely found in California.

Findings from this study confirmed that the *Anagrus* wasps in Yolo County, and *A. daanei* in particular, will successfully attack the *E. ziczac* from Mendocino and Lake County. The fact that *E. ziczac* “source population” had an effect on parasitism rates may indicate that the *E. ziczac* in Yolo County have some localized adaptation that improves their ability to defend against *Anagrus* parasitoids or, alternately, that the *Anagrus* wasps in Yolo County have greater preference for *E. ziczac* hosts from Mendocino and Lake County. Further studies would be needed to more accurately evaluate this observation.

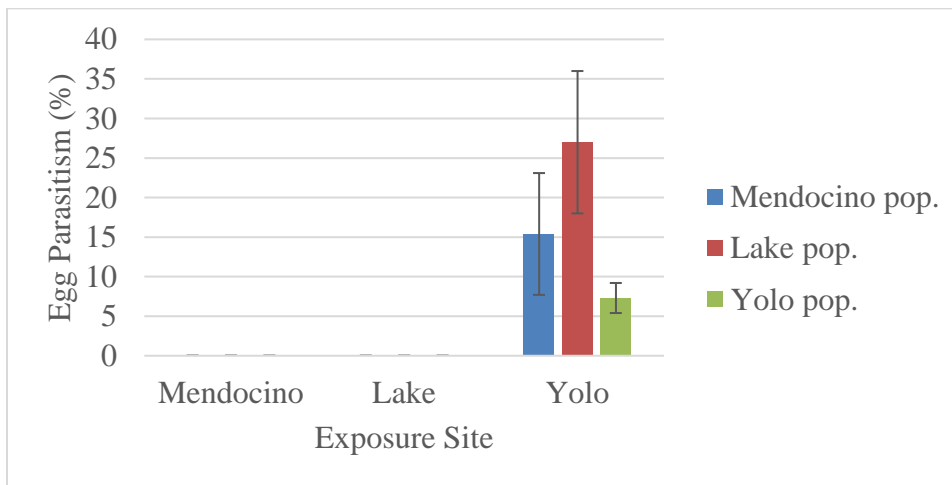


Figure 7. Parasitism of *E. ziczac* eggs from Mendocino, Lake and Yolo County.

Table 3. Results of the statistical analysis

Variable	Likelihood Ratio Test ( $\chi^2$ )	P-value
Exposure site	465.83	<0.001
Source population	64.01	<0.001
Total eggs per vine	3.53	0.06

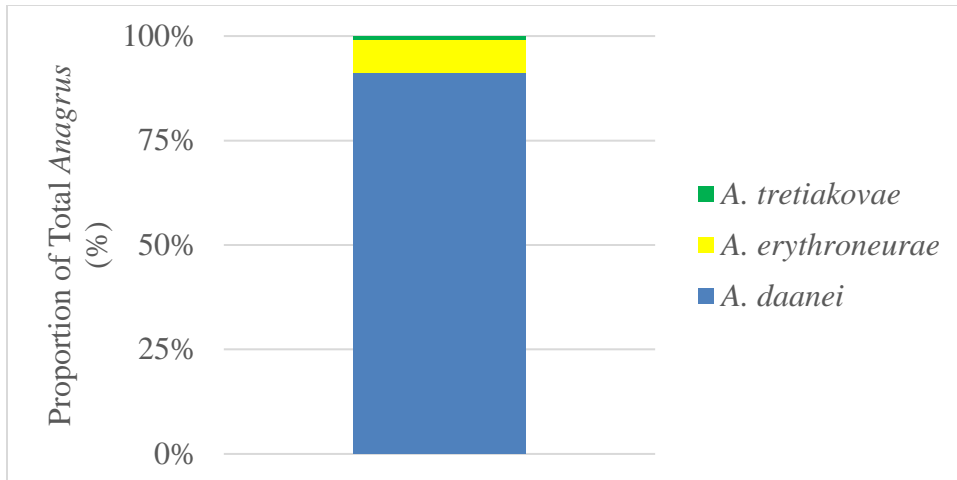


Figure 8. Species composition of the *Anagrus* wasps attacking *E. ziczac* in Yolo County.

## **7. Outside Presentations of Research**

Oral Presentations (22 total):

“New and Old Pest and Disease in the Vineyard.” CAPCA Continued Education Seminar - California Association of Pest Control Advisers. Napa, CA. 45 people in attendance. Feb. 2014 ***Presented by Dr. Lucia Varela***

“Update on a leafhopper, a bug and a moth” Sonoma County Grape Day - UCCE Sonoma County. Santa Rosa, CA. 220 people in attendance. Feb. 2014 ***Presented by Dr. Lucia Varela***

“The importance of *Anagrus* wasps for biological control of grape leafhoppers in California” Virginia Creeper Leafhopper Seminar - UCCE North Coast. Ukiah, CA. Mar. 2014 ***Presented by Dr. Houston Wilson***

“Updates on vineyard pests: mites, leafhoppers, moths, other arthropods or exotic pests” Sonoma County Winegrape Commission Pest Control Adviser Breakfast Meetings - Sonoma County Winegrape Commission. Windsor, CA. 100 people in attendance. Mar. 2014 ***Presented by Dr. Lucia Varela***

“Arthropod pests in the vineyard, identification, life cycle and control for vineyard supervisors and farmworkers” 2014 Napa Valley Grapegrowers Farmworker Education Workshop - Napa Valley Grapegrowers. Napa, CA. 150 people in attendance. Mar. 2014 ***Presented by Dr. Lucia Varela***

“Monitoring and control measures updates of pests of pears and grapevines” Mendocino Pest Control Advisers Breakfast Meeting. Ukiah, CA. 36 people in attendance. Mar. 2014 ***Presented by Dr. Lucia Varela***

“Biology and Control of Virginia creeper leafhopper in Mendocino and Lake Counties” Virginia Creeper Leafhopper Seminar - UCCE Mendocino County. Ukiah, CA. 65 people in attendance. Mar. 2014 ***Presented by Dr. Lucia Varela***

“Bio-Control in a pesticide based IPM program” CAPCA – ED Seminar. San Ramon, CA. Mar. 2014. ***Presented by Dr. Kent Daane***

“The role of generalist predators and parasitoids in grape leafhopper control in California.” Virginia Creeper Leafhopper Seminar, Ukiah, CA. Mar. 2014. ***Presented by Dr. Kent Daane***

“IPM Practices in Viticulture” Santa Rosa Junior College Integrated Pest Management Class - Santa Rosa Junior College. Forestville, CA. 30 people in attendance. May 2014 ***Presented by Dr. Lucia Varela***

“Insecticide trial results presentation and field evaluation of damage” Virginia creeper leafhopper Field Meeting. Hopland, CA. 25 people in attendance. June 2014 ***Presented by Dr. Lucia Varela***

“Vineyard Pest Identification and Monitoring of Key Insect Pests for Vineyard Workers” Employee Development Workshop - Sonoma County Winegrape Commission. Santa Rosa, CA. 45 people in attendance. July 2014 ***Presented by Dr. Lucia Varela***

“Viral pathogens in vineyards: What do we really know about insects and the dispersal of red blotch?” Oakville Winegrowers Association. Oakville, CA. July 2014 ***Presented by Dr. Kent Daane***

“Variegated grape leafhopper in California vineyards - why a key invasive pest has lost its pest status.” British Columbia Wine Grape Council Annual Meeting. Pendicton, Canada. July 2014 ***Presented by Dr. Kent Daane***

“The importance of insects as vectors of grape leafroll associated viruses: a California perspective” British Columbia Wine Grape Council Annual Meeting. Pendicton, Canada. July 2014 ***Presented by Dr. Kent Daane***

“Hands-on insect identification demonstration: leafhoppers, sharpshooters, stinkbugs, moths” Sonoma Winegrape Commission Sustainable Day - Sonoma Winegrape Commission. Forestville, CA. 90 people in attendance. Aug. 2014 ***Presented by Dr. Lucia Varela***

“California grape pests, field tour.” Agro-Kanesho Study Group (Japanese Ag-Business). Kearney Agricultural Research and Extension Center. Parlier, CA. Sept. 2014. ***Presented by Dr. Kent Daane***

“Biological control of Virginia creeper leafhopper” Lake and Mendocino County IPM Seminar - UCCE Lake and Mendocino County. Lakeport, CA. Nov. 2014 ***Presented by Dr. Houston Wilson***

“Biological control of the Virginia creeper leafhopper (*Erythroneura ziczac*)” Entomology Society of America Annual Meeting. Portland, OR. Nov. 2014 ***Presented by Dr. Houston Wilson***

“Improving biological control of the Virginia creeper leafhopper (*Erythroneura ziczac*)” Current Issues in Vineyard Health. Davis, CA. Dec. 2014 ***Presented by Dr. Houston Wilson***

“Improving biological control of the Virginia creeper leafhopper (*Erythroneura ziczac*)” UC ANR Grape Workgroup Meeting. Davis, CA. Dec. 2014 ***Presented by Dr. Houston Wilson***

“An overview of leafhopper biology and management in vineyards.” Current Issues in Vineyard Health. Davis, CA. Dec. 2014. *Presented by Dr. Kent Daane*

Publications (2 total):

**Varela, L. G. & K. M. Daane.** 2013. Virginia creeper leafhopper. In *Grape Pest Management Manual*. L.J. Bettiga (Ed.) 3rd ed. Oakland: Univ. Calif. Div. Agric. Nat. Res. Publ. 3343. Chapter 36: 235-236.

**Varela, L.G. & G. McGourty.** 2014. Leafhopper control using OMRI-approved insecticides. Sonoma County UCCE publication 5 pp.

### **Strategy for Communicating Results to End-Users:**

As this work progresses, we will continue to present relevant findings and updates on *E. ziczac* management to a variety of grower and industry stakeholders. Venues would likely include grower and professional society meetings (i.e. Allied Grape Growers, County Farm Bureau, Association of Applied Insect Ecologists etc.). We have already made plans for a grower-researcher meeting to be held in Mendocino and Lake County sometime during the week of February 16-20, 2015. This meeting will mark the beginning of our work to coordinate an area-wide IPM program to promote grower adoption of best management practices for Virginia creeper leafhopper control. Research results will also be published in both scientific and industry journals (i.e. Environmental Entomology, Practical Vineyard and Winery, UC ANR publications etc.). Information will also be made available through county extension websites.

### **8. Research Success Statements**

We evaluated three OMRI-approved insecticides and found that two applications of Stylet oil or DeBug® Turbo targeting younger nymphs of the first brood were as effective at controlling *E. ziczac* populations as one Pyganic® treatment. Based on this information, growers can now consider oil treatments as a viable alternative to repeated use of Pyganic®, which may lead to resistance. Applications of any OMRI product for the control of *E. ziczac* must be put on earlier in the season (i.e. during the first brood) due to their increased fecundity relative to *E. elegantula*, which many growers are able to tolerate until the second or third brood.

Vineyard surveys revealed that parasitism of *E. ziczac* is relatively non-existent in many areas, including Mendocino, Lake, Napa (Pope Valley) and El Dorado County. In contrast, *E. ziczac* populations in Yolo County were regularly parasitized by *A. daanei*. Surprisingly, we did find *A. daanei* in vineyards, but it was only attacking *E. elegantula*, whereas in Yolo County *A. daanei* was attacking both *E. ziczac* and *E. elegantula*. The identification of an *A. daanei* population that attacks *E. ziczac* was the first step in improving control of this pest in the other regions of northern California where growers are currently experiencing severe outbreaks, in large part due to a total lack of biological control.

Molecular studies of *A. daanei* is still in progress, but preliminary morphological assessments have shown that the *A. daanei* in Yolo and Mendocino County are not separate species. At present, it appears that *A. daanei* is failing to attack *E. ziczac* in the regions where

this pest is a relatively new arrival. Our hypothesis is that the *A. daanei* in these areas have been reproducing solely on *E. elegantula* for so long that they have effectively lost their preference for *E. ziczac* even though this is a viable host for this *Anagrus* species. Improving our knowledge of *Anagrus* systematics improves our ability to correctly identify and manipulate these parasitoids to improve biological control of *E. ziczac* in commercial vineyards.

We attempted to force *A. daanei* from the Mendocino County population to attack *E. ziczac* eggs in order to know whether or not they would accept this alternate host in the absence of their preferred host, *E. elegantula*. With hundreds of *A. daanei* enclosed with potted grape vines containing hundreds of *E. ziczac* eggs there was no instance where the eggs were ever attacked by this *Anagrus* species. These findings rule out the possibility that *A. daanei* populations found in the North Coast region might rapidly adapt to *E. ziczac*.

Finally, we successfully demonstrated that the *A. daanei* population in Yolo County could attack and reproduce on *E. ziczac* eggs from Mendocino and Lake County. This is very promising news for growers in those regions where there is effectively zero parasitism of this pest (i.e. Mendocino, Lake, Napa and El Dorado County). Now that we have confirmed that the Yolo County population of *A. daanei* will readily attack *E. ziczac* from other regions, it may be possible for us to improve biological control in other regions by introducing these wasps into commercial vineyards.

## **9. Funds Status**

All funds are being appropriately spent and we foresee that funds will be completely used by the end of the granting period. Salary positions include partial funding for a post-doctoral researcher (Dr. Houston Wilson). Houston was responsible for carrying out the northern California survey work and conducting the experiments with *Anagrus* parasitoids. Serguei Triapitsyn identified the *Anagrus* specimens to species and coordinated the molecular work at UC Riverside. Lucia Varela conducted the spray trial in Mendocino County. Glenn McGourty and the Mendocino County extension office assisted the grant by providing their lab tech Ryan Keiffer to help Lucia and Houston with field work. Kent Daane provided guidance for experimental design and laboratory space at the UC Berkeley Oxford Tract greenhouse. Travel costs include trips to field sites in Napa, Sonoma, Mendocino, El Dorado and Yolo County. Supplies and expenses costs were primarily used for mounting, identification and molecular work with *Anagrus* specimens and field supplies for the survey and *Anagrus* experimental work.

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